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## The species of the genus *Diamesa* (Diptera, Chironomidae) known to occur in Italian Alps and Apennines

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### Abstract

Some rare species from Italian Alps, belonging to the genus *Diamesa* Meigen, 1835 (Diptera, Chironomidae) are here re-described as adult males, because only old, incomplete descriptions are available for these taxa. Terminology of male genitalia is reviewed, diagnostic features are illustrated in detail, and notes on biology and geographical distribution of the examined species are provided. An identification key to the known adult males is presented.

**Key words:** Diamesinae, Diamesini, taxonomy, aquatic insects, male genitalia

### Introduction

New conservation policies for freshwater ecosystems need to be developed as a priority due to the presence of rare species with high conservation value. Growing threats to their existence require attention, considering their importance in maintaining natural food webs (Moore *et al.* 2009). Effective conservation strategies require the appropriate knowledge on the morphology of the rare species as well as an update of their distribution and ecology. Global warming threatens cold-stenothermal species (Marziali & Rossaro 2013; Soszyńska-Maj *et al.* 2015) and in particular the glacial retreat seriously endangers species living near the glacier snout. Insect communities of this habitat are dominated by Chironomidae (Diptera) belonging to the genus *Diamesa* Meigen, 1835 (Diptera, Chironomidae). These species are at risk of disappearance due to the global warming occurred in the last decades. Unfortunately, an updated list of *Diamesa* species inhabiting the Italian Alps and Apennines is currently missing, as it is from many other mountain ranges in the world where the chironomid fauna is completely unknown. Despite both the rarity of these species and the threat to their existence, descriptions of some species are still based on outdated papers.

The present work re-describes the males of some rare species of the genus *Diamesa* Meigen, 1835 collected in glacial habitat. In the Italian Alps, 21 species belonging to the genus *Diamesa* were recorded since the 1970's, including four rare species that were collected from one or two sites at most. The two rare species represented by *D. martae* Kownacki & Kownacka, 1980 and *D. nowickiana* Kownacki & Kownacka, 1975 are here redescribed, their diagnostic features are illustrated in detail, and notes on biology and geographical distribution are provided. The other two, *D. longipes* Goetghebuer, 1941 and *D. wuelkeri* Serra-Tosio, 1964 were collected in poor conservation state, so a detailed re-description is not possible here. Attention is devoted to male genitalia and a discussion of the terminology used by different authors in genitalia description is presented. A key to adult males of *Diamesa* from the Italian Alps is also provided.

## Material and methods

The material examined was mounted on slides following the procedure outlined by Wirth & Marston (1968) and Sæther (1969); briefly boiling the specimen in KOH 10 %, transferring it in acetic acid and finally clearing it in a phenol: xylene mixture 3:1 before its mounting in Balsam.

Measurements were made using a LEICA DM LS B2 optic microscope at different magnifications (40 – 1000) connected to a LEICA DFC320 camera. Drawings were prepared with a drawing tube connected to the optic microscope. Measurements are given in µm or in mm as specified.

The examined specimens were collected in 331 sites near the actual glaciers across Italy; a complete list of the sampling localities is reported in Rossaro & Lencioni (2015a, b).

The specimens are deposited in the Dipartimento di Scienze per gli Alimenti, la Nutrizione e l'Ambiente (DeFENS – Università degli Studi di Milano) except *D. martae* and one specimen of *D. nowickiana*, which are deposited in Museo delle Scienze (Muse – Trento).

## Results

### Notes on the nomenclature of male genitalia of the genus *Diamesa*

There are some still opened questions about the nomenclature of different parts of male genitalia. The morphological nomenclature of male genitalia in *Diamesa* is based on contributions of different authors (Serra-Tosio 1964; 1968; 1970; 1971a, b; 1972; Hansen & Cook 1976; Sæther 1980; Willassen 2005) (Table 1). The French terms 1—*pièce médiane*, 2—*partie externe* of *pièce dorsale* and 3—*partie interne* of *pièce dorsale* (Serra-Tosio 1971b) coincide with the English/Greek terms 1—*sternapodeme*, 2—*phallapodeme* and 3—*aedeagal lobe* (Fig. 1) respectively (Hansen & Cook 1976; Sæther 1980). The French terms open the question of the correspondence with the English terms used by the other authors. The French terms *appendice supérieur* and *appendice inférieur* were used describing the species belonging to *Diamesa latitarsis* group (Serra-Tosio 1964; 1968). The *appendice supérieur* is here named dorso-lateral lobe of inferior volsella and coincides with the dorso-medial finger-like projection in Hansen & Cook (1976), while the *appendice inférieur* is here named ventro-medial lobe of inferior volsella and coincides with the medial spine and seta bearing projection described in Hansen & Cook (1976); an accurate analysis emphasizes that it is divided into two parts (Fig. 2). In *D. lavillei* (Serra-Tosio 1969) the *appendice supéro-antérieur* and the *appendice supéro-postérieur* are here named superior and inferior volsella, respectively; while *appendice inférieur* clearly coincides with the lobe bearing the basimedial setal cluster. In *D. veletensis* (Serra-Tosio 1971a) the *appendice antéro-latéral* refers to the superior volsella, the *postéro-lateral* to the inferior volsella, while the *petite appendage* coincides with the lobe bearing the basimedial setal cluster. In *D. vaillanti* (Serra-Tosio 1972) the *appendice antéro-latéral* is again the superior volsella, while the *appendice medio-lateral* and *postéro-lateral* correspond with the two lobes in which inferior volsella is divided. The identification of basal plate and median field (Hansen & Cook 1976) with superior and inferior volsella respectively (Sæther 1980) is not followed by Willassen (2005) in the description of *D. solhoysi*. This last author named superior volsella a third appendage visible between basal plate and median field, emphasizing that superior volsella and basal plate are not the same structure. In the present paper the identification of basal plate with superior volsella and of median field with inferior volsella are maintained (Hansen & Cook 1976; Sæther 1980). This conclusion is supported by the observation that the basal plate in *Diamesa* is in the same position of superior volsella described in all Orthocladiinae (Cranston *et al.* 1989), and by the observation that in *Diamesa* the median field is often divided into a dorsal and a ventral lobe, which well corresponds to the dorsal and ventral parts of inferior volsella observed in *Orthocladius* (Rossaro *et al.* 2003; Sæther 2005). In any case, the matter requires further investigation. The terminology of hypopygial appendages used in the present study and correspondence with that the one previously used by different authors is given in Table 1.

**TABLE 1.** Terminology of hypopygial appendages according to different authors and the terminology proposed here (first column).

Present paper	Saether 1980	Hansen & Cook 1976	Serra-Tosio 1964, 1968, 1971b
sternapodeme	sternapodeme	sternapodeme	pièce médiane
phallapodeme	phallapodeme	phallapodeme	partie externe of pièce dorsale
aedeagal lobe	aedeagal lobe	aedeagal lobe	partie interne of pièce dorsale
superior volsella	superior volsella	basal plate	
inferior volsella	inferior volsella	median field	
dorso-lateral lobe of inferior volsella		dorso-medial finger-like projection	appendice supérieur
ventro-medial lobe of inferior volsella		medial spine and seta bearing projection	appendice inférieur
basimedial setal cluster		basimedial setal cluster	

continued.

Present paper	Serra-Tosio 1969	Serra-Tosio 1971a	Serra-Tosio 1972	Willlassen 2005
sternapodeme				sternapodeme
phallapodeme				
aedeagal lobe				aedeagal lobe
superior volsella	appendice supéro- antérieur	appendice antéro-lateral	appendice antéro- lateral	basal plate
inferior volsella	appendice supéro- postérieur	appendice postéro-lateral		median field
dorso-lateral lobe of inferior volsella			appendice medio- lateral	superior volsella ?
ventro-medial lobe of inferior volsella			appendice postéro- lateral	
basimedial setal cluster	appendice inférieur	petite appendage		basimedial setal cluster

## Notes and redescriptions

### *Diamesa dampfi* (Kieffer, 1924)

(Fig. 1 A, B)

The adult males of the species can be easily identified based on the shape of the dark, well chitinized aedeagal lobe bearing 2–3 stout spines medially (Fig. 1 A, B). The species was described and illustrated in Pagast (1947) and Serra-Tosio (1970). *Diamesa dampfi* was collected in cold springs above 1000 m of altitude (Rossaro & Lencioni 2015a, b).

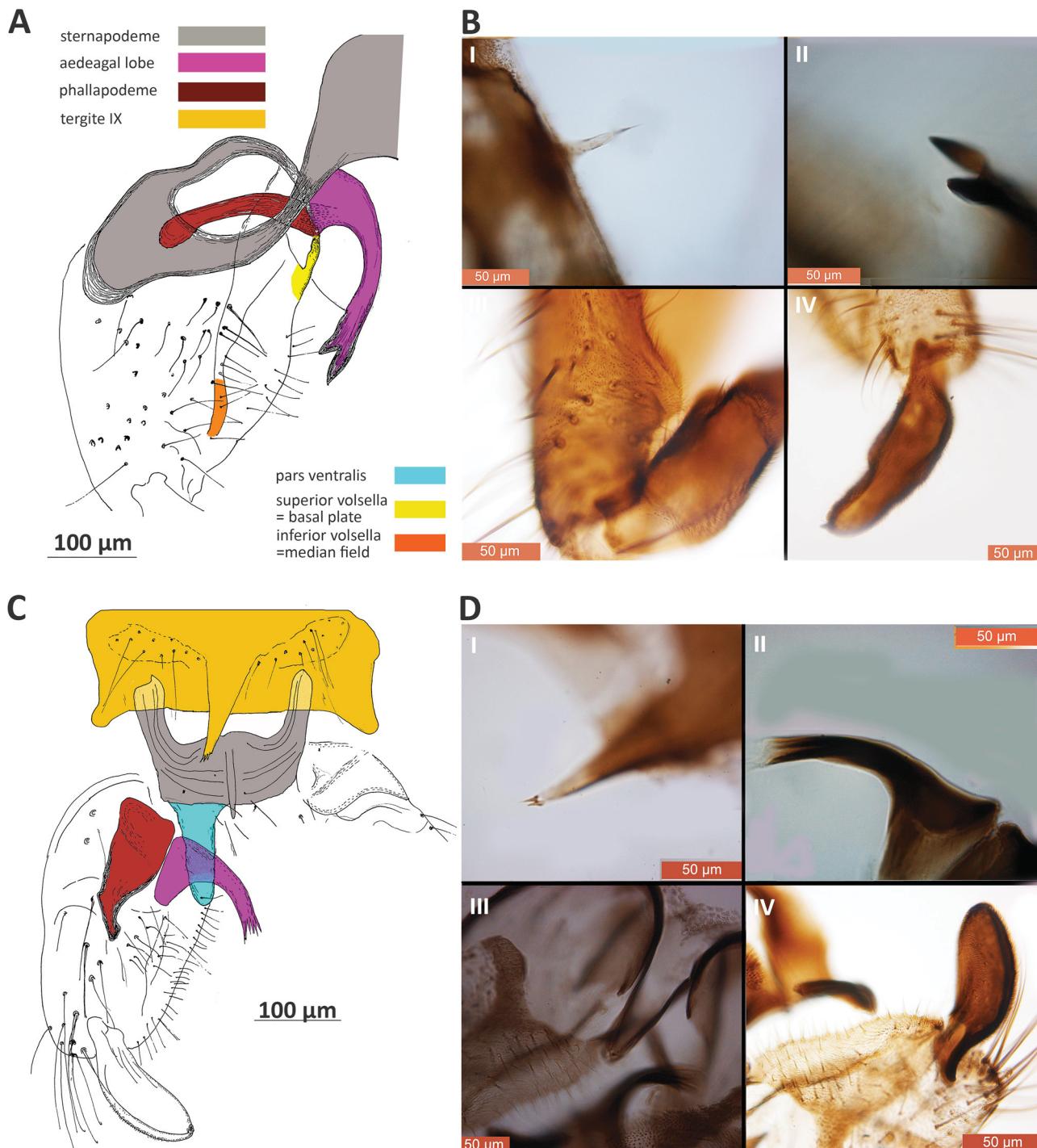
### *Diamesa permakra* (Walker, 1856)

(Fig. 1 C, D)

The species can be easily identified on the basis of the shape of the aedeagal lobe bearing only fine spines at apex (Fig. 1 C, D). There is an old drawing of this species (Pagast 1947), and the most recent ones are in Serra-Tosio (1971b). In the paper with the original description of *D. thomasi* Serra-Tosio, 1970 (the *dampfi* group), some other diagnostic details are also provided.

In Italy, only an adult male associated to pupal exuviae was collected in Acqualba stream in 1978 (Prealps,

NO). Larvae and pupal exuviae cannot be separated from *D. dampfi*; the larvae and pupae recorded in few localities in Prealp springs (Rossaro & Lencioni 2015a, b) probably belong to *D. permacra*.



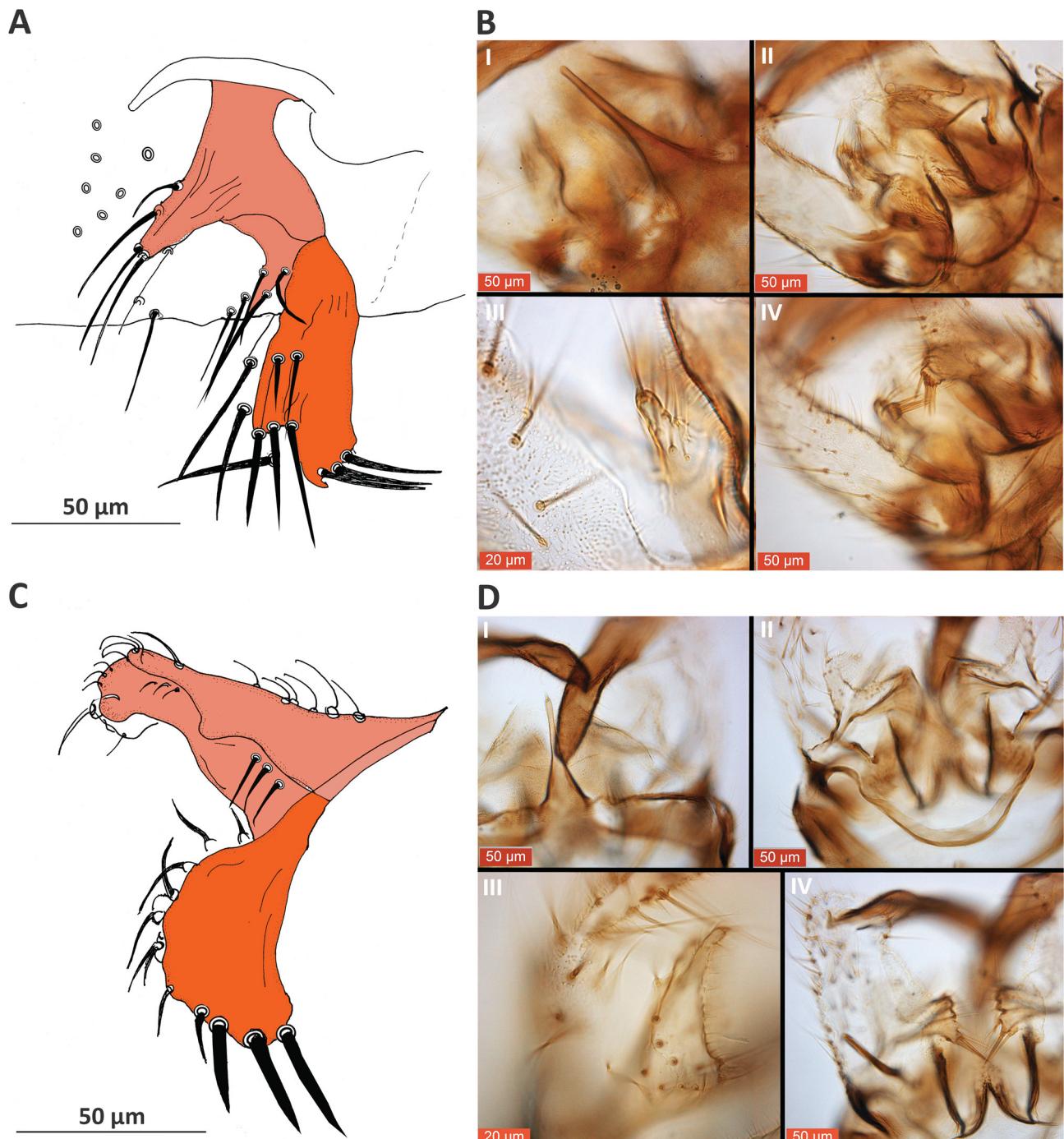
**FIGURE 1.** **A**, *Diamesa dampfi* (Kieffer), male genitalia and legend of colors used to mark the different parts; **B I**, anal point, **B II**, aedeagal lobe, **B III**, inferior volsella, **B IV**, gonocoxite; **C**, *Diamesa permacra* (Walker), male genitalia; **D I**, anal point, **D II**, aedeagal lobe, **D III**, pars ventralis, **D IV**, gonocoxite.

#### *Diamesa latitarsis* (Goetghebuer, 1921)

The species is well described by Serra-Tosio (1968), with notes on the variability found on dorso-lateral and ventro-medial lobes of inferior volsella. Beside the Alps, where it is relatively common, an adult male was recorded in the Apennines (Rossaro & Lencioni 2015a, b).

*Diamesa modesta* Serra-Tosio, 1968

The species publication year is here accepted to be 1968 following Ashe & O'Connor (2009). The species is well described by Serra-Tosio (1968) and can be separated from *D. latitarsis* based on the larger and shorter inferior volsella with dorso-lateral and ventro-medial lobes almost fused. It is known from different localities in the Alps (Rossaro & Lencioni 2015a, b).



**FIGURE 2.** **A**, *Diamesa lindrothi* Goetghebuer in Goetghebuer & Lindroth, inferior volsella with dorso-lateral and ventro-medial lobes; **B I**, anal point, **B II**, male genitalia, **B III**, inferior volsella dorso-lateral lobe, **B IV**, inferior volsella ventro-medial lobe; **C**, *Diamesa goetghebueri* Pagast, inferior volsella with dorso-lateral and ventro-medial lobe; **D I**, anal point, **D II**, male genitalia, **D III**, inferior volsella dorso-lateral lobe, **D IV**, inferior volsella ventro-medial lobe.

***Diamesa lindrothi* Goetghebuer in Goetghebuer & Lindroth, 1931**

(Fig. 2 A, B)

The examination of genitalia of many adult males suggests that in the Alps there are transition forms between this species and *D. goetghebueri*. The species *D. lindrothi* has a digitiform dorso-lateral lobe of inferior volsella (Fig. 2 A, B), while in *D. goetghebueri* the lobe is larger and flat (Fig. 2 C, D). According to Serra-Tosio (1973) this species is distributed throughout the Scandinavian Peninsula, but it is also present in the Alps. Examining larvae with barcode analysis confirms the existence of at least two separated species under this name in the Alps (Montagna *et al.* 2016).

***Diamesa goetghebueri* Pagast, 1947**

(Fig. 2 C, D)

The species is not easily distinguished from *D. lindrothi* (see above). It could be suspected that the digitiform (*D. lindrothi*) or flat (*D. goetghebueri*) aspect of dorso-lateral lobe of inferior volsella (Fig. 2) depends on the angle of visualization. However, an accurate examination of many specimens from Estellette and Adamello glacial streams, with the male genitalia rigorously mounted in the same dorsal position, confirms the presence both of a digitiform and a flat dorso-lateral lobe and supports the separation of two species (see figures). According to Serra-Tosio (1973), *D. goetghebueri* is endemic from the Alps (Rossaro & Lencioni 2015a, b).

***Diamesa wuelkeri* Serra-Tosio, 1964**

(Fig. 3 A, B)

This species is endemic from the Alps (Serra-Tosio 1964; 1968). In Italy, an adult male of this species was collected only in Viola stream in July 1985 (Rossaro & Lencioni 2015a, b). For details of male genitalia see Fig. 3 A, B. The species can be diagnosed based on the ventro-medial lobe of inferior volsella (“appendice inférieur”) that is much better developed than the much reduced dorso-lateral lobe.

***Diamesa laticauda* Serra-Tosio, 1964**

The species is very rare in the Alps. It is recorded only in few localities: Estellette glacial stream, Veny Valley (Val d’Aosta), 11IX1995, 12IX1997; Lys Glacial stream 30VI1987; Vedretta de la Mare (Adamello) 31VII2002. It can be easily distinguished by the expansion of the gonostylus and the large pars ventralis (Serra-Tosio 1964).

***Diamesa longipes* Goetghebuer, 1941**

(Fig. 3 C, D)

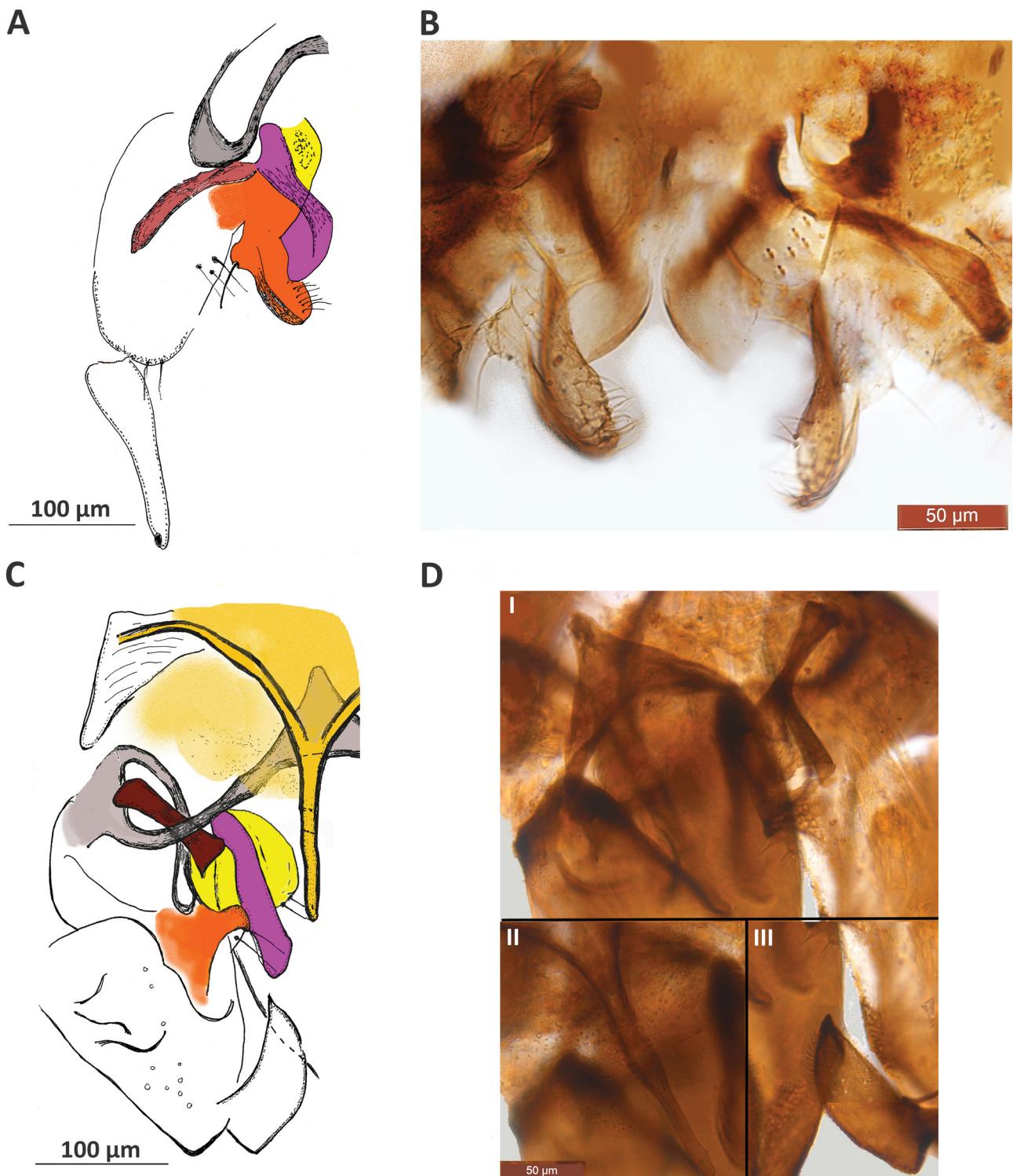
The species was redescribed and figured by Kownacka & Kownacki (1975), and the male genitalia are here redrawn (Fig. 3 C, D). The beak-like shape of inferior volsella is typical. The species was found in the Italian Alps at 12VIII1978 near the Avio Lake, and no more recorded thereafter.

***Diamesa martae* Kownacki & Kownacka, 1980**

(Fig. 4 A, B)

Some additional information to the original description are here given (the measurements given in the original description are in parenthesis).

Adult male: body length 4.7 (6.5 mm), wing length 3.62 (4.4 mm), general color of the body dark brown.



**FIGURE 3.** **A**, *Diamesa wuelkeri* Serra-Tosio, male genitalia; **B**, dorso-lateral and ventro-medial lobes of inferior volsella; **C**, *Diamesa longipes* Goetghebuer, male genitalia; **D I**, sternapodeme, **D II**, anal point, **D III**, gonostylus.

Antenna with 13 flagellomeres, non-plumose; each flagellomere with very short setae (160 µm); pedicel spherical, on its lateral side the group of 4 setae described by Kownacki & Kownacka (1980) is not visible. Eyes bare. Flagellomeres length-width ratios: (120:52, 40:40, 48:40, 40:32, 40:32, 36:28, 36:24, 40:28, 48:28, 44:28, 52:24, 52:24, 616:40), pedicel (80:140). AR (1.04).

Maxillary palp four-segmented; the length-width ratio of each segment in µm: 86.6:58, 129:52, 158:43, 188:40 (72:60, 200:60, 172:40, 220:40).

Legs: see Table 2.

**TABLE 2.** Lengths (in  $\mu\text{m}$ ) and proportions of leg segments. (Legends: KP<sub>I,II,III</sub>: measurements given in the original description, IP<sub>I,II,III</sub>: measurements of the specimen from the Italian Alps; fe, femur; ti, tibia; ta1–5, tarsomeres 1–5; LR, leg ratio; BV, combined length of femur, tibia, and ta1 divided by combined length of tarsomeres 2–5; SV, ratio of femur plus tibia to ta1.)

	fe	ti	ta1	ta2	ta3	ta4	ta5	LR	BV	SV
KP <sub>I</sub>	2380	2513	1530	714	459	170	209	0.60	4.16	3.20
KP <sub>II</sub>	2635	2295	1037	554	340	170	204	0.45	4.71	4.75
KP <sub>III</sub>	2771	2686	1530	782	425	170	204	0.57	4.42	3.57
IP <sub>I</sub>	1844	1965	1230	630	370	112	148	0.63	4.00	3.10
IP <sub>II</sub>	1924	1582	927	457	275	100	130	0.59	4.61	3.78
IP <sub>III</sub>	2059	2167	1631	940	366	102	134	0.75	3.80	2.59

Hypopygium (Fig. 4 A, B; Fig. 3 in Kownacki & Kownacka 1980): tergite IX divided into two parts by a less chitinized surface passing into a short, poorly chitinized anal point, two groups of setae situated on brighter areas on the tergite symmetrically to the body axis; gonocoxite with two fairly characteristic appendages: superior volsella rounded, not clearly differentiated, inferior volsella with two lobes, an anterior dorsal lobe digitiform, well separated from gonocoxite, projecting medially, and a posterior ventral lobe elongated, adhering to gonocoxite; outer surface of the gonocoxite covered with longer hairs; two strong setae on the postero-median margin of gonocoxite; gonostylus black-brown, slender, somewhat swollen in median part, ending with apical spine, covered wholly with sensilla, with short setae on bright areas. Transverse sternapodeme with a shape intermediate between the typical arc (as in many *Diamesa* species, e.g. *D. zernyi*) and the triangular shape characterizing species of the *steinboecki* group; the hook-shaped aedeagal lobe is characteristic.

Adult female, pupa, larva: unknown.

**Material collected:** One male from Amola glacier stream, 2474 m a.s.l., 14VIII2014.

**Systematic remarks.** According to Kownacki & Kownacka (1980) the determination of a group membership of this species is difficult. It belongs most probably to the *Diamesa latitarsis* group. The bare eyes and the structure of the hypopygium provide arguments for including it here. The gonocoxite inferior volsella is composed by two lobes, as in all other species of *latitarsis* group, but the shape of these lobes is characteristic in this species. The presence of two strong setae on the postero-median margin of gonocoxite and the shape of gonostylus also suggests the inclusion in the *latitarsis* group, whereas the sternapodeme resembles the one observed in *D. steinboecki*.

#### *Diamesa nowickiana* Kownacki & Kownacka, 1975

(Fig. 4 C, D)

Additions to the original description (Kownacki & Kownacka 1975) are here provided (in parenthesis are reported the measures of the original description).

Adult male: length 4.8 mm (4.0 mm), wing length 3.674 mm (2.5 mm), wing width 1 mm, coloration brownish black.

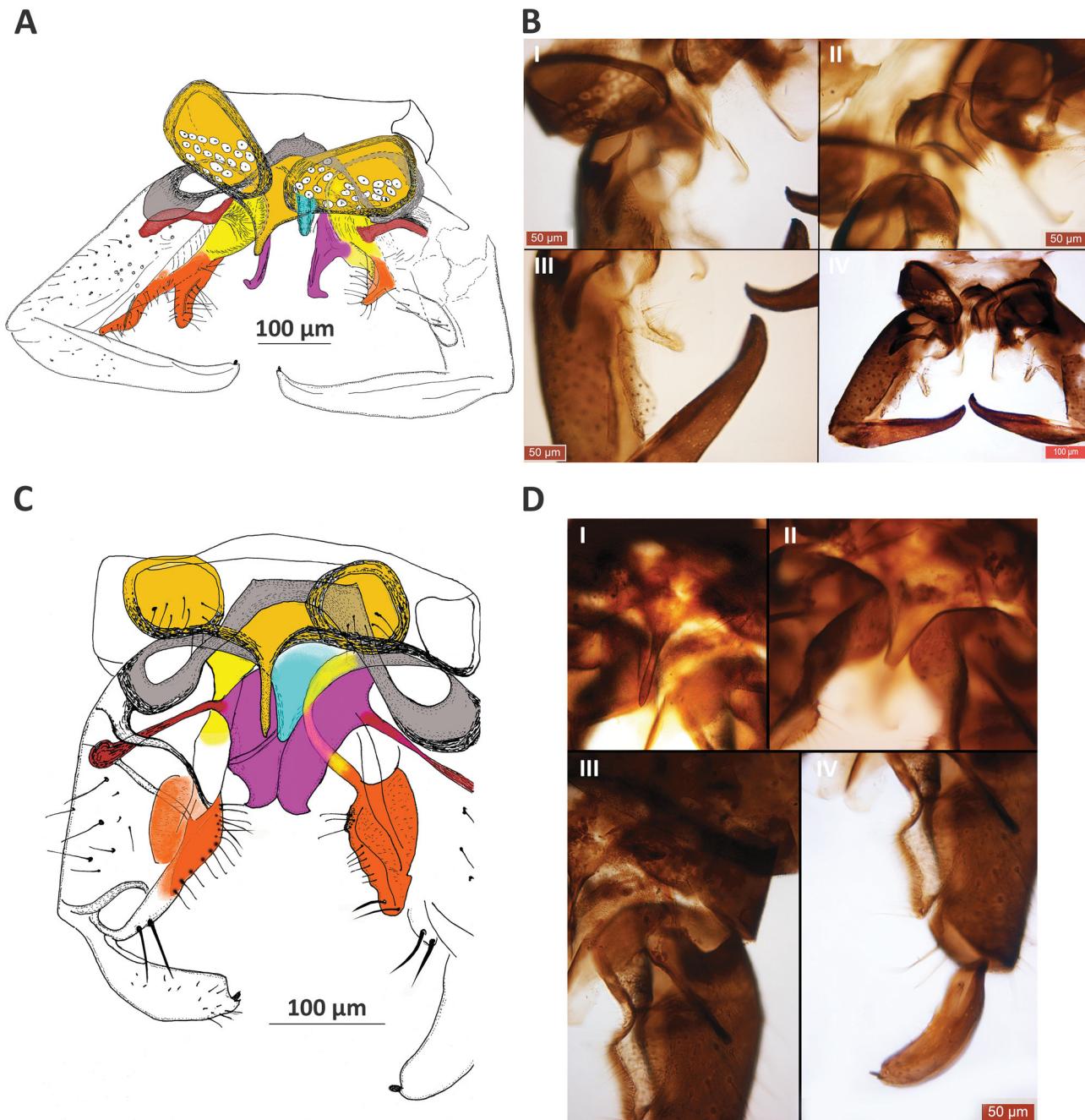
Head dark brown, eyes haired, the hairs longer than ommatid, the head chaetotaxy as for the other species of the genus *Diamesa*. Antenna lost in the Amola specimen, in holotype with thirteen segments (pedicel: 120:72, 20:24, 28:28, 28:28, 32:40, 40:40, 44:476  $\mu\text{m}$ ), AR (0.87–0.92), all segments with bristles. Palp shorter than antenna, consisting of four segments: not measureable: 116: 158: 164  $\mu\text{m}$  (80:128:112:160).

Hypopygium (Fig. 4 C, D): Anal point short and wide, rounded apically, without bristles. IX abdominal segment with two groups of dorsal hairs symmetrically placed along the body line. Pars ventralis large. Aedeagal lobe characteristic, with a point in postero-lateral position. Gonocoxite with a superior volsella not projecting, almost square in shape, gonocoxite slightly concave in the middle part with a densely pubescent inferior volsella, enlarged in the middle, with two robust setae apically. Two strong setae in postero-median corner of gonocoxite.

Gonostylus widening towards the end, apical spine shorter than the back edge of the gonostylus, with well-developed sclerotized structures inside.

**Systematic remarks.** According to Kownacki & Kownacka (1980) the species belongs to the *aberrata* group, but the presence of hairy eyes suggests an affinity with the *bertrami* group. *Diamesa nowickiana* is easily separated from all the other species by the shape of the anal point.

**Material collected:** One pharate pupa with genitalia visible, Presena glacial stream, 13IX1990; one male, Amola glacier 14VIII2014.



**FIGURE 4.** **A**, *Diamesa martae* Kownacki & Kownacka, male genitalia; **B I**, anal point, **B II**, sternapodeme and pars ventralis, **B III**, inferior volsella, **B IV**, gonostylus; **C**, *Diamesa nowickiana* Kownacki & Kownacka, male genitalia; **D I**, anal point, **D II**, pars ventralis, **D III**, phallapodeme and inferior volsella, **D IV**, inferior volsella and gonostylus.

### ***Diamesa bertrami* Edwards, 1935**

The species is redescribed and illustrated in the unpublished thesis of Serra-Tosio (1971b). The species is coldstenothermal (Marziali & Rossaro 2013) and one of the commonest species in glacial habitat (Rossaro & Lencioni 2015a, b).

### ***Diamesa aberrata* Lundbeck, 1898**

The most recent illustrations of male genitalia of this species are given in the thesis of Serra-Tosio (1971b), in Hansen & Cook (1976) and in Makarchenko (1985). Pagast (1947) described but did not illustrated the species. The species inhabits glacial streams as well as cold springs and streams (Rossaro & Lencioni 2015a, b).

### ***Diamesa incallida* (Walker 1856)**

Illustrations and descriptions of male genitalia of this species are by Pagast (1947), Serra-Tosio (1971b), Hansen & Cook (1976), and Makarchenko (1985). The species is restricted to cold springs (Rossaro & Lencioni 2015a, b).

### ***Diamesa starmachi* Kownacki & Kownacka, 1970**

The most recent illustrations of male genitalia of this species are in Gilka *et al.* (2013). The species was collected in the Italian Alps in Vermigliano stream and in Noce Bianco, but only in the larval and pupal stages (Rossaro & Lencioni 2015a, b). No adult male of *D. starmachi* are known at present from Italy.

### ***Diamesa zernyi* Edwards, 1933**

(Fig. 5 A, B)

The most recent illustrations of male genitalia of this species are in Serra-Tosio (1971b) and in Serra-Tosio (1972). The male genitalia are here redrawn (Fig. 5 A, B). The species is common in glacial habitat, in different microhabitats (Rossaro & Lencioni 2015a, b).

### ***Diamesa vaillanti* Serra-Tosio, 1972**

The species is well described and illustrated by Serra-Tosio (1972). It is very near to *D. zernyi*, but can be distinguished by the shorter and larger gonocoxite and inferior volsella (Serra-Tosio 1972). The species inhabits glacial streams, and it is widespread in cold waters at high altitudes (Rossaro & Lencioni 2015a, b).

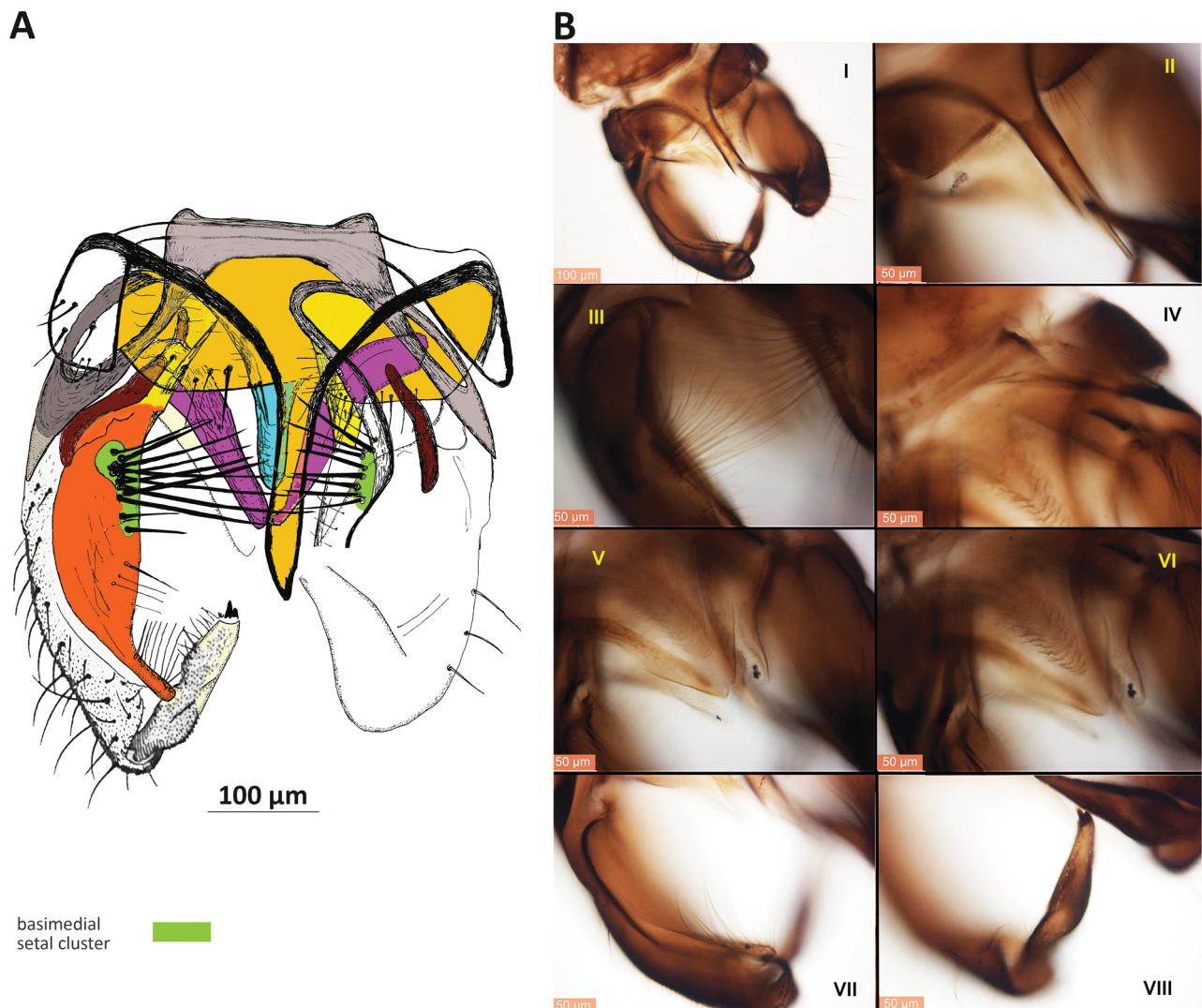
### ***Diamesa tonsa* (Haliday in Walker, 1856)**

The illustrations of male genitalia of this species are by Pagast (1947) (as *D. thienemanni*) and Serra-Tosio (1971b). The species is the most common and widespread *Diamesa* species in the Alps, and is also common in the Apennines (Rossaro & Lencioni 2015a, b).

### ***Diamesa cinerella* (Meigen in Gisti, 1835)**

The species is redescribed by Pagast (1947) and Serra-Tosio (1971b). Species delimitation analysis performed with Cytochrome c oxidase subunit I (Montagna *et al.* 2016) suggested that the species is close to *D. tonsa*, but the adult

males are morphologically well separable to each other. The species is common in glacial streams but in the winter it is also present at low altitudes, as in Ticino River (Rossaro & Lencioni 2015a, b).



**FIGURE 5.** **A**, *Diamesa zernyi* Edwards, male genitalia (basimedial setal cluster in green); **B I**, IX tergite, **B II**, anal point, **B III**, basimedial setal cluster, **B IV**, sternapodeme and phallapodeme, **B V**, aedeagal lobe, **B VI**, pars ventralis, **B VII**, inferior volsella, **B VIII**, gonostylus.

#### *Diamesa insignipes* Kieffer in Kieffer & Thienemann, 1908

(Fig. 6 A, B)

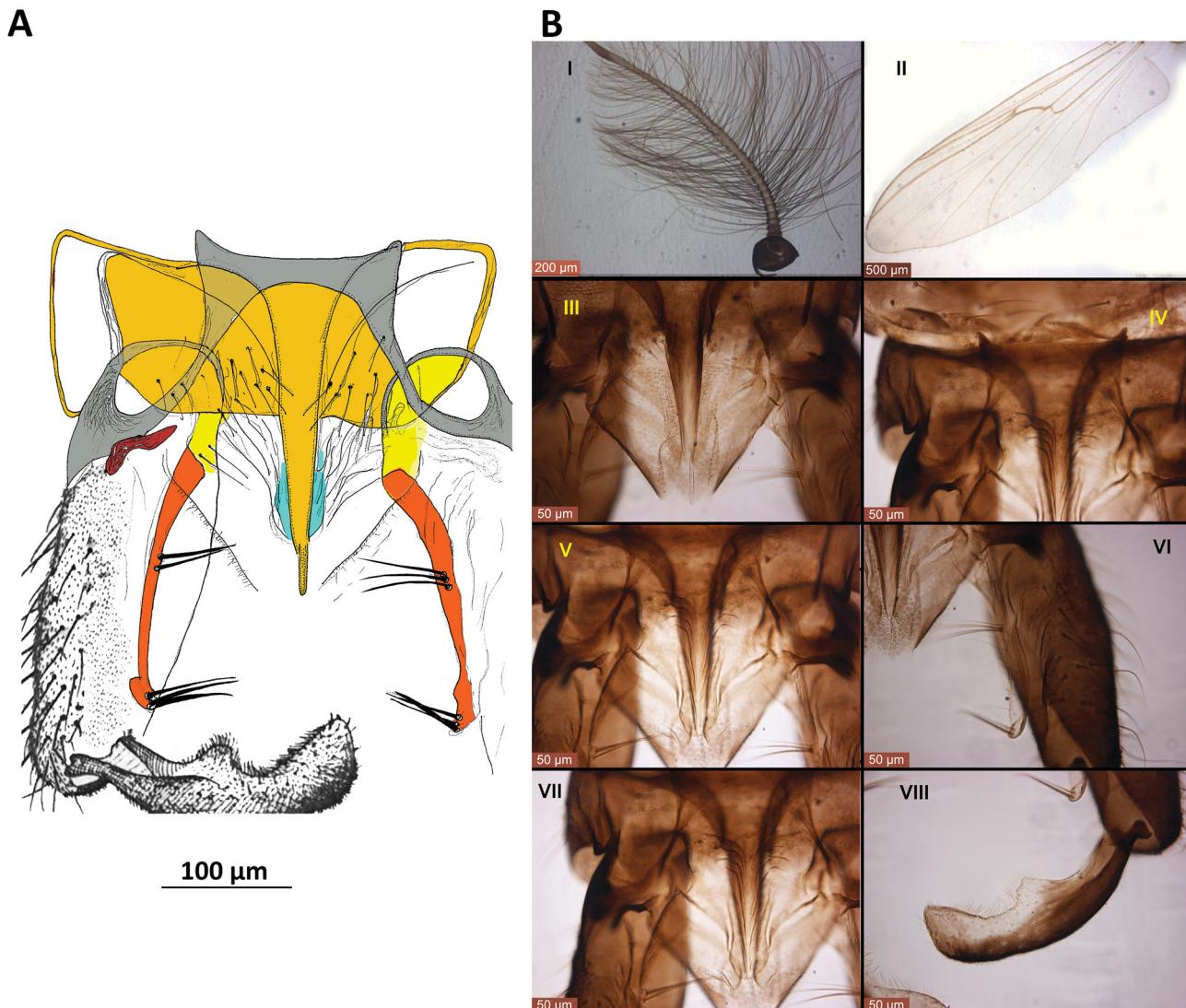
Illustrations of male genitalia of this species are reported by Pagast (1947), Serra-Tosio (1971b), Hansen & Cook (1976), and Makarchenko (1985). Hitherto, the species was never recorded in the Alps, whereas in Prealps the species was found in Pioverna stream only (long E 9.38, lat N 46.00). It is noteworthy that males of the Pioverna population have the inferior volsella bearing more than two robust bristles. The species inhabits also the following Apennines streams: Nure, Taro, Ceno, and Sangro (Rossaro & Lencioni 2015a, b).

#### Key to species of *Diamesa* from the Italian Alps based on adult males

- 1 Antenna plumose, with 13 flagellomeres; sternapodeme arched; eyes bare or hairy; gonostylus of variable shape, never triangular or squared; wings never reduced; microtrichia on medial surface of gonostylus less dense ..... 2
- Antenna not plumose with 8 flagellomeres; sternapodeme triangular; eyes hairy; gonostylus triangular large at its base and

	restricted to apex or squared; wings often reduced; medial surface of gonostylus with "pile" of microtrichia . . . . .	19
2	Eyes hairy, that is the eye microtrichia clearly surpass the ommatidial lens and are visible along lateral eye margin . . . . .	3
-	Eyes not hairy, that is the eye microtrichia are not visible or at least not surpass the height of ommatidial lenses along lateral eye margin . . . . .	9
3	Basimedial setal cluster present, in ventral position, between superior and inferior volsella. . . . .	4
-	Basimedial setal cluster absent . . . . .	7
4	Superior volsella flat, not well separable from the rest of gonocoxite; gonocoxite more or less elongated, slender; basimedial setal cluster with about 12–15 very long setae, directed medially; inferior volsella elongated, with numerous long (60–70 µm), anteriorly directed setae on the distal part; gonostylus uniform without some part enlarged. . . . .	5
-	Superior volsella rounded, well developed, clearly separable from the rest of the gonocoxite; gonocoxite not unusually long and slender; basimedial setal cluster present but with very short setae; inferior volsella not elongated, without numerous anteriorly directed setae; gonostylus with a bulbous basal part and a slender distal part about of the whole length . . . . .	6
5	Gonocoxite long and slender (length 400–500 µm, breadth 70–80 µm); inferior volsella slender (200 µm long, 20 µm wide) with a long digitiform projection, directed caudally . . . . .	<i>D. zernyi</i>
-	Gonocoxite shorter and wider (300–400 µm, breadth 160–180 µm); inferior volsella stouter (200 µm long, 60–70 µm wide), digitiform projection of inferior volsella short . . . . .	<i>D. vaillanti</i>
6	Inferior volsella simple appearing as a large flap; gonostylus with the slender distal part longer than the proximal bulbous part . . . . .	<i>D. cinerella</i>
-	Inferior volsella double with lobes of different length, the lateral one longer; gonostylus with the slender distal part as long as the bulbous proximal part . . . . .	<i>D. tonsa</i>
7	Gonostylus very long, about 0.7 times length of gonocoxite; inferior volsella long, slender, with 1–3 long medially directed setae at about 0.5 its length and about 3 long setae at its distal end . . . . .	<i>D. insignipes</i>
-	Gonostylus short, less than 0.7 times the length of gonocoxite; inferior volsella short, expanded medially, with many strong setae ventro-medially . . . . .	<i>bertrami</i> group ... 8
8	Anal point present, but short, with rounded apex, not projecting beyond T <sub>IX</sub> ; inferior volsella adhering to gonocoxite without robust setae; aedeagal lobes large triangular, with a characteristic point in postero-lateral position; two robust setae on the postero-medial corner of gonocoxite . . . . .	<i>D. nowickiana</i>
-	Anal point very long, well projecting beyond T <sub>IX</sub> ; inferior volsella not adhering to gonocoxite, with strong setae directed medially; aedeagal lobes without a characteristic point in postero-lateral position; no robust setae on postero-medial corner of gonocoxite . . . . .	<i>D. bertrami</i>
9	Inferior volsella present, but not well separated from gonocoxite . . . . .	16
-	Gonocoxite with a well separated inferior volsella . . . . .	10
10	Sternapodeme with a pointed median part; inferior volsella divided in two lobes, but only medial lobe well separated from gonocoxite; gonocoxite without two robust setae in its distal part . . . . .	<i>D. martae</i>
-	Sternapodeme arched; inferior volsella divided into two or three parts, all well separated from gonocoxite; gonocoxite with two robust setae in its distal part . . . . .	<i>latitarsis</i> group 11
11	Gonostylus without a strong median expansion; anal point long and slender; pars ventralis digitiform, elongated . . . . .	12
-	Gonostylus with a median expansion; anal point short and large; pars ventralis very large . . . . .	<i>D. laticauda</i>
12	Inferior volsella with a small, dorso-lateral flat or finger-like projection and two stronger, ventro-medial spine- and seta-bearing projections . . . . .	13
-	Inferior volsella with dorso-lateral lobe very reduced or elongated, but never digitiform or flat, ventro-medial lobe without spine or seta bearing projections . . . . .	14
13	Dorso-lateral lobe of inferior volsella finger-like short but well visible . . . . .	<i>D. lindrothi</i>
-	Dorso-lateral lobe of inferior volsella, large flat . . . . .	<i>D. goetghebueri</i>
14	Dorso-lateral lobe very small difficult to see; ventro-medial lobe of inferior volsella very developed, elongated, bearing spoon-like supple setae . . . . .	<i>D. wuelkeri</i>
-	Dorso-lateral lobe of inferior volsella well developed, longer than the ventro-medial lobe . . . . .	15
15	Dorso-lateral and ventro-medial lobes of inferior volsella well separated to each other; dorso-lateral lobe more elongated, slender . . . . .	<i>D. latitarsis</i>
-	Dorso-lateral and ventro-medial lobes of inferior volsella almost fused; dorso-lateral lobe less elongated, only a little longer than ventro-medial lobe, wider . . . . .	<i>D. modesta</i>
16	Anal point absent; pars ventralis short, T <sub>IX</sub> very weak medially, appearing to be formed of two separate sclerites; inferior volsella flat, densely covered of setae, barely protruding from gonocoxite . . . . .	<i>D. incallida</i>
-	Anal point present; pars ventralis and T <sub>IX</sub> well developed; inferior volsella never protruding from gonocoxite . . . . .	17
17	Eyes apparently bare, but with very short pubescence visible at large magnification; inferior volsella present, with slightly expanded distal end and moderately long setae; anal point short, shorter than the length of T <sub>IX</sub> ; aedeagal lobes normal; tarsomeres cordiform; gonocoxite without lobes; gonostylus fairly slender, straight, of nearly equal width throughout . . .	<i>D. aberrata</i>
-	Eyes completely devoid of microtrichia; inferior volsella very weakly differentiated, without sharp dorsal margin and without expanded distal end; anal point needle-like, not quite or just reaching distal ends of aedeagal lobes; aedeagal lobes very sclerotized, dark; tarsomeres cylindrical; gonocoxite with a small lobe in caudal position, gonostylus fairly sharply narrowed basally . . . . .	18
18	Aedeagal lobes, with 4–5 fine apical denticles; anal point larger, at least 10 µm wide, with two small apical spines; gonostylus without an enlargement in the middle . . . . .	<i>D. permacra</i>
-	Aedeagal lobes, with two large apical spines; anal point less than 10 µm wide, with only one apical spine; gonostylus with an	

- enlargement in the middle ..... *D. dampfi*
- 19 Anal point short, weak, directed ventrally; phallapodeme short, narrow, appearing as a lateral projection of aedeagal lobe; inferior volsella weak, only outlined without free distal end; gonostylus triangular ..... 20
- Anal point very long, basal part sclerotized, distal part not sclerotized; phallapodeme wide, robust; inferior volsella beak-like, with a group of long setae ventrally; gonostylus almost squared ..... *D. longipes*
- 20 Anal point short and slender (75–120 µm); inferior volsella visible only as a small appendage; total body length 2.0–3.5 mm ..... *D. steinboecki*
- Anal point short, triangular; gonocoxite without visible inferior volsella; total body length 3.2–3.4 mm ..... *D. starmachi* (known only as larvae and pupal exuviae from the Italian Alps)



**FIGURE 6.** **A**, *Diamesa insignipes* Kieffer in K. & Thienemann, male genitalia; **B I**, antenna, **B II**, wing, **B III**, anal point, **B IV**, IX tergite and superior volsella, **B V**, aedeagal lobe, **B VI**, inferior volsella, **B VII**, pars ventralis, **B VIII**, gonostylus.

## Discussion

The review of the Palearctic *Diamesa* species known to occur as adult males and pupae (Pagast, 1947) was followed by many contributions describing species from the Alps (Serra-Tosio 1964; 1968; 1970; 1971a, b; 1972; 1983); in the meanwhile a review of Nearctic species (Hansen & Cook 1976) was added; other contributions from other Holarctic regions were added (Makarchenko 1985). A recent complete list of worldwide *Diamesa* species is in Ashe & O'Connor (2009), but a re-description of all the known species is outside the aim of the present work. Larvae of many species were recently redescribed (Rossaro & Lencioni 2015a, b), but there are still many gaps in larval knowledge. In the present paper the adult males of some rare species collected in the Italian Alps are

redrawn, terminology of genitalia is reviewed, hoping that these re-descriptions will aid in the identification of species that will be collected in the future.

Only 12 of the 21 species of *Diamesa* recorded until now in the Italian Alps and Apennines are known in all developmental stages (semaphoronts). At present, the larvae belonging to the *D. latitarsis*, *tonsa*, *cinerella* and *zernyi* groups can be separated to each other, although different species within the same group cannot be distinguished. These are the species with the broadest distribution in the Italian Alps. The association of larvae with adults is possible examining reared material or mature pupae collected in the field with associated larval exuviae, but this is actually available only for these relatively abundant species. The description of all semaphoronts is much more critical for rare species. At present, *D. longipes*, *D. martae* and *D. nowickiana* are known only as adult males, while *D. starmachi* is known from Italy only as larva and prepupa. It is difficult, even impossible, to identify species from larval material using morphological characters alone, so larval association to a well identifiable species is often problematic (Rossaro & Lencioni 2015a, b). Innovative strategies of specimens identification based on molecular markers represent promising tools to solve the previously reported issue, but it has been developed only on a limited number of species (Montagna *et al.* 2016).

The interest focused on *Diamesa* species is bound to their serious risk of extinction, being inhabitants of cold waters, recently undergoing towards a dramatic warming especially in the periphery of their areas of distribution, as in the Southern Alps, and the captures of *Diamesa* species are becoming rarer in recent years, because of the increasing temperatures and the glaciers retreat (Rossaro *et al.* 2016). Since all the discussed species live in cold waters of glacial or high mountain streams, the consequence of the glacial retreat is the further isolation of their populations in the short-term and, in the long-term, their possible disappearance due to very restrict habitat preference and limited migration abilities.

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